

ORIGINAL ARTICLE

Effect of freezing, hot tumble drying and washing with eucalyptus oil on house dust mites in soft toys

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Abstract

Soft toys are a major source of house dust mites (HDM) and HDM allergens, and sleeping with soft toys is a significant risk factor for HDM sensitization. We studied three techniques to eliminate HDM from soft toys, namely freezing, hot tumble drying and washing with eucalyptus oil. Thirty-six toys (12 in each treatment group) were enumerated for live HDM by the heat escape method before and after freezing overnight, hot tumble drying for 1 h and washing in 0.2% to 0.4% eucalyptus oil. Freezing, hot tumble drying and washing with eucalyptus oil resulted in significant reductions in live HDM, an average reduction of 95.1%, 89.1% and 95.1%, respectively. Additionally, washing with eucalyptus oil resulted in a significant reduction in HDM allergens as well from a geometric mean of 9.12 µg/g to 0.37 µg/g ($p = 0.033$). These three HDM elimination techniques give parents of infants effective and acceptable methods of limiting HDM exposure.

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House dust mite (HDM) allergens induce IgE-mediated sensitization in susceptible individuals and can cause asthma symptoms in those sensitized (1, 2). As sensitization to HDM allergens is associated with the development of asthma in children (3), that the severity of asthma symptoms and HDM exposure in the indoor environment shows a dose-response relationship (4) and that HDM allergen avoidance can reduce asthma symptoms (5), the presence of HDM and HDM allergens in soft toys could be of importance for children who routinely sleep with them.

Soft toys are a major source of HDM allergens (6), and at a very early age (0–2 yr), sleeping with soft toys is strongly associated with HDM sensitization (7). It is generally recommended that HDM-sensitized asthmatics practice allergen avoidance. To be effective, HDM allergen avoidance methods have to target both HDM and their allergens. Both chemical and physical techniques have been used. These include the use of acaricides, tannic acid, eucalyptus oil and physical techniques such as freezing and hot tumble drying.

It has been suggested that if soft toys are frozen overnight to -20°C , it will kill HDM. Similarly, eucalyptus oil treatment of clothing is acaricidal to HDM (8) and hot tumble drying kills HDM in bedding (9). The aim of our study was to determine

whether overnight freezing of soft toys, hot tumble drying or adding eucalyptus oil to the wash cycle kills HDM. To our knowledge, the use of eucalyptus oil and hot tumble drying has not previously been used to kill HDM in soft toys.

Methods

Participants

We approached mothers at Changhua Christian Hospital with children who attended the Hospital's child care facility and asked them whether they could bring in one of their child's soft toys for study. Thus, 12 mothers were recruited in each of the three studies (freezing, hot tumble drying and eucalyptus oil wash) resulting in a total of 36 soft toys. The study protocols were explained and informed written consent obtained. The study was approved by the Changhua Christian Hospital Ethics Committee.

Freezing study

Initially, the soft toys were analysed for the presence and number of live HDM by the heat escape method as previ-

ously described (9). Briefly, five strips of adhesive tape, 50×1.9 cm (total area: 0.0525 m^2) were applied to one side of the soft toy and heat directed at the opposite side for 2 h with a 250-W heat lamp suspended 35 cm above the site. Live HDM were then counted on the adhesive strips under $20\times$ magnification with a stereomicroscope. Mites were considered live if full bodied and moving, and dead if inactive, even when touched with a blunt probe. The soft toys were then placed in a domestic deep freezer at about -15°C overnight (at least 16 h). The next morning, the soft toys were allowed to reach room temperature and live HDM measured on the opposite side of the soft toys as described previously.

Hot tumble drying study

The soft toys were analysed for live HDM before and after hot tumble drying as described previously. The soft toys were individually tumble dried for 1 h on the maximum setting in a domestic clothes dryer (Panasonic, Taiwan). After drying and before analysing for live HDM, the soft toys were placed in a humid environment (average relative humidity: 66.7%; range: 60–72%) overnight to allow any stunned, but live HDM to recover.

Eucalyptus oil washing

As the eucalyptus oil had to be added to the washing water with the use of liquid detergents and not all commercial brands of liquid detergents are able to emulsify homogeneously, we initially tested a number of liquid detergents. To test whether the liquid detergent was suitable, we used the following criteria (8):

- the eucalyptus oil should dissolve and form a clear solution at a ratio of one part detergent to 3–5 parts of oil;
- when a teaspoon of the emulsion is mixed with about 200 ml of water, a milky, opaque solution that is stable (does not break down or separate) for at least 10 min should form.

We found one brand of liquid detergent that satisfied the above criteria (Magic Amah detergent).

Before washing, live HDM were measured by the heat escape method as described previously. The soft toys were then totally submersed in an aqueous solution of the eucalyptus oil/detergent emulsion so that the final concentration of the eucalyptus oil in the wash solution was between 0.2% and 0.4%. The soft toys were left in this solution for 1 h with occasional stirring and then removed, thoroughly rinsed under tap water and then dried indoors for 48 h. Live HDM numbers were then measured as described previously. Additionally, one side of the soft toy was vacuumed for 1 min and dust collected for allergen analysis into nylon mesh bags as previously described (9), both before and after the washing procedure.

House dust mite allergens analysis

One hundred milligram aliquots of dust were extracted with 1 ml of phosphate-buffered saline at room temperature and

extracts stored at -4°C before allergen analysis in one analytical batch. The HDM allergens Der p 1 and Der f 1 are both prevalent in Changhua City, Taiwan (10) and therefore both allergens were measured in the extracts by double-monoclonal antibody ELISA methods using commercial kits (Indoor Biotechnologies, Charlottesville, VA, USA) as previously described (11).

Statistical analysis and data presentation

Life HDM numbers are presented as mean numbers per m^2 with range of values and total HDM allergen (Der p 1 + Der f 1) concentrations as geometric means $\mu\text{g/g}$ with range of values. Comparison of these before and after treatment (freezing, hot tumble drying and washing) was made by the exact Wilcoxon test for nonparametric paired data with statistical significance set at the p 0.05 level.

Results

All 36 soft toys (average weight: 145 g; range: 25–270 g) contained significant numbers of live HDM before treatment (range: 96–5269 HDM/ m^2). Table 1 shows that freezing, hot tumble drying and washing with eucalyptus oil were all effective in greatly reducing the number of live HDM.

Washing with eucalyptus oil was effective in significantly reducing HDM allergens (Der p 1 + Der f 1) in 12 soft toys from a geometric mean of $9.12 \mu\text{g/g}$ (range: 0.42–147.9) to $0.37 \mu\text{g/g}$ (range: 0.01–5.3; $p = 0.033$).

Discussion

Soft toys are a known source of HDM allergens (6, 12), and it has recently been shown that sleeping with soft toys is associated with an increased rate of sensitization to aeroallergens with an odds ratio of 1.45 (7). As most children are emotionally attached to their toys and frequently sleep with them in close proximity of their airways, ways of reducing allergen exposure from toys is highly desirable.

Washing bedding and clothing items in cold or warm water is known to reduce HDM allergens, but has no effect on live HDM (13). A wash temperature of at least 55°C is required to kill HDM, and this is not a practical way to eliminate live HDM from soft toys as this temperature may destroy the fabric. Tovey and McDonald subsequently showed that by pre-soaking woollen blankets for 30 min in a

Table 1 Effect of freezing, hot tumble drying and washing with eucalyptus oil on house dust mites (HDM) in soft toys

	Life HDM/ m^2 Before	Life HDM/ m^2 After	% Reduction
Freezing	693 (206–1970)	17* (0–106)	95.1 (51.4–100)
Drying	1341 (96–5269)	209** (0–1199)	89.1 (62.1–100)
Washing	1108 (193–2837)	18*** (0–106)	95.1 (45.0–100)

Results are means with range of values.

* $p = 0.0004$; ** $p = 0.010$; *** $p = 0.0007$.

warm detergent solution (30°C) with 0.2% eucalyptus oil was 97% effective in killing HDM. We have shown that this procedure is also effective in killing HDM in soft toys with an average 95.1% reduction in live HDM. Of the 12 soft toys studied, 10 showed no live HDM after washing with eucalyptus oil, one showed a reduction in live HDM of 96.4% and one only showed a reduction of 45%. With regard to this soft toy showing only a 45% reduction in live HDM, it was the smallest toy at 25 g weight and had the lowest number of live HDM pre-wash (193/m²). Washing is known to reduce HDM allergen levels significantly, and this was also evident in our study where the geometric mean levels of Der p 1 + Der f 1 were reduced from 9.12 µg/g pre-wash to 0.37 µg/g post-wash.

Hot tumble drying for 1 h in a domestic clothes dryer has been shown to be effective in substantially reducing live HDM in duvets, but does not result in a reduction in HDM allergen levels (9). In that study, duvets contained a mean number of live HDM of 410/m² before tumble drying and this was significantly reduced to a mean number of 6/m². We have shown that this technique is also significantly effective in killing HDM in soft toys. The number of live HDM in toys (mean: 1341/m²) was much higher than in the duvets study, and we achieved an average reduction in live HDM of 89.1%. Some of the soft toys only showed a moderate reduction in live HDM; this may be because of the thickness of toys affecting the maximum temperature reached inside the toy. The thermal death point of *Dermatophagoides pteronyssinus* is 55°C (14), and domestic clothes dryers can achieve a maximum temperature inside duvets greater than 55°C and a relative humidity of less than 10% during a 1-h drying period (9). A limitation of our study is that we did not measure the temperature inside the soft toys during tumble drying; however, we achieved a similar HDM killing rate as our previous study with duvets.

Popular advice to parents of infants is to put soft toys in a domestic deep freezer overnight to kill HDM, and a Japanese

laboratory-based study achieved an almost 100% mortality when live HDM were frozen at -20°C for 30 min (15). Our study has shown that placing soft toys in a domestic deep freezer at -15°C for at least 16 h was quite effective in killing HDM, although there was some variability most likely due to the thicker material of some of the toys.

A limitation of our study was that we did not determine how quickly the soft toys were recolonized with HDM. A previous study has demonstrated that after application of liquid nitrogen to carpets, virtually all HDM were killed, but recolonization of HDM occurred within 1 month (16). Also, in a study of placing new, allergen-free, pillows on domestic beds, accumulation of the major HDM allergen, Der p 1, was rapid (17). Thus, soft toys are likely to be rapidly recolonized with HDM and this requires further studies so that practical advice can be given on how often the toys need to be treated.

Washing with eucalyptus oil results in the soft toys smelling of eucalyptus oil post-wash. This is generally pleasant and acceptable. However, it is not known whether this is a potential problem in patients with bronchial hyperresponsiveness and may be a problem for some infants. We are not aware of any reported cases of eucalyptus oil-induced exacerbations of asthma symptoms at the concentrations used in our study.

In conclusion, we have shown that three commonly used techniques, freezing, hot tumble drying and washing with eucalyptus oil, are equally effective in killing HDM in soft toys. Of the three techniques, washing with eucalyptus oil also has the benefit of greatly reducing HDM allergens concentrations as demonstrated by Tovey and McDonald (8). However, simple washing techniques could also be applied after freezing or hot tumble drying to reduce HDM allergens levels. These three techniques give parents of infants effective and acceptable methods of limiting HDM exposure from soft toys.

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